PTO/SB/68 (11-95)
Approved for use through 10/31/99. CMB 0651-0031
Patent and Trademark Office: U.S. DEPARTMENT OF COMMERCE
Patent and Trademark Office: U.S. DEPARTMENT OF COMMERCE
is the Paperwork Reduction Act of 1995, no persons are recurred to respond to a collection of information unless it displays a valid OMB control number.

	In re Application of	f .	•	•
	·		•	
	Application Number	er	Filed	
	1 1			
	08/876	.276		
	Group Art Unit	Examiner		· ·
			,	2
				223
			aper No	12
			aper 110	
Assistant Commissioner for Patents				3
Vashington, DC 20231				0:0
hereby request access under 37 CFR 1				
hereby request access under 37 C. N. S. Jentified ABANDONED application, which were to in United States Pate (B) referred to in an application that Application No.	nt Number 6 /	cot f	omb in 37 CF	R 1.11. I.E
	filed			
Application No	•		. :	
Application No paper number,	,		ion that is 00	en to public
Application No, paper number, (C) an application that claims the being pertion, i.e., Application No.	enefit of the filing care	of an applicat	ion that is op	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No (D) an application in which the application to the public.	enefit of the filing cate	of an applicat	ion that is op filed y open the co	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No. (D) an application in which the application	enefit of the filing cate	of an applicat	ion that is op filed y open the co	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No (D) an application in which the application to the public.	enefit of the filing cate	of an applicat	ion that is op filed y open the co	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No (D) an application in which the application to the public.	enefit of the filing cate	of an applicat	ion that is op filed y open the co	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No (D) an application in which the application to the public.	enefit of the filing cate	cf an applicat	ion that is op filed y open the co address:	en to public , or
Application No	enefit of the filing cate	cf an applicat	ion that is op filed y open the co	en to public , or
Application No, paper number, (C) an application that claims the be inspection, i.e., Application No (D) an application in which the application to the public.	enefit of the filing cate	cf an applicat	ion that is op filed y open the co address:	en to public, or omplete
Application No. paper number	enefit of the filing cate	cf an applicat	ion that is op filed y open the co address:	en to public, or or omplete





(12) United States Patent Short et al.

(10) Patent No.:

US 6,174,673 B1

(45) Date of Patent:

*Jan. 16, 2001

(54) HIGH THROUGHPUT SCREENING FOR NOVEL ENZYMES

(75) Inventors: Jay M. Short, Encinitas; Martin Keller, San Diego, both of CA (US)

(73) Assignee: Diversa Corporation, San Diego, CA

(US)

(*) Notice: Under 35 U.S.C. 154(b), the term of this patent shall be extended for 0 days.

This patent is subject to a terminal dis-

(21) Appl. No.: 09/098,206

(22) Filed: Jun. 16, 1998

Related U.S. Application Data

(63)	Continuation-in-part of application No. 08/876,276, filed on Jun. 16, 1997.

(51)	Int. Cl. ⁷		C12Q	1/68
------	-----------------------	--	------	------

(52)	U.S. Cl.	
		435/471 · 435/476 · 435/320 1

(56) References Cited

U.S. PATENT DOCUMENTS

FOREIGN PATENT DOCUMENTS

WO 98/56904 12/1998 (WO). WO 99/49315 9/1999 (WO). WO99/54494 10/1999 (WO).

OTHER PUBLICATIONS

Zhang et al. FASEB J. vol. 5, pp. 3108-3113, 1991.*

Plovins et al. Applied and Environmental Microbiology. vol. 60(12), pp. 4638-4641, 1994.*

* cited by examiner

Primary Examiner—Remy Yucel
(74) Attorney, Agent, or Firm—Gray Gary Ware &
Freidenrich LLP; Lisa A. Haile

(57) ABSTRACT

Disclosed is a process for identifying clones having a specified activity of interest, which process comprises (i) generating one or more expression libraries derived from nuclei acid directly isolated from the environment; and (ii) screening said libraries utilizing a fluorescence activated cell sorter to identify said clones. More particularly, this is a process for identifying clones having a specified activity of interest by (i) generating one or more expression libraries derived from nucleic acid directly or indirectly isolated from the environment; (ii) exposing said libraries to a particular substrate or substrates of interest; and (iii) screening said exposed libraries utilizing a fluorescence activated cell sorter to identify clones which react with the substrate or substrates. Also provided is a process for identifying clones having a specified activity of interest by (i) generating one or more expression libraries derived from nucleic acid directly or indirectly isolated from the environment; and (ii) screening said exposed libraries utilizing an assay requiring co-encapsulation, a binding event or the covalent modification of a target, and a fluorescence activated cell sorter to identify positive clones.

23 Claims, 16 Drawing Sheets